

A Synthetic Antagonist for the Peroxisome Proliferator-activated Receptor γ Inhibits Adipocyte Differentiation*

(Received for publication, September 10, 1999)

Harold M. Wright^{‡§}, Clary B. Clish[¶], Toshiyuki Mikami^{||}, Stefanie Hauser^{‡ **}, Kazunori Yanagi^{||},
Ryuji Hiramatsu^{||}, Charles N. Serhan[¶], and Bruce M. Spiegelman^{‡ ‡‡}

From the [‡]Dana-Farber Cancer Institute and Department of Cell Biology, Harvard Medical School, [¶]Center for Experimental Therapeutics and Reperfusion Injury, Department of Anesthesia, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts 02115, and ^{||}Biotechnology Laboratory, Sumitomo Chemical Company, Takarazuka 665-8555, Japan

While searching for natural ligands for the peroxisome proliferator-activated receptor (PPAR) γ , we identified a synthetic compound that binds to this receptor. Bisphenol A diglycidyl ether (BADGE) is a ligand for PPAR γ with a K_d (app) of 100 μ M. This compound has no apparent ability to activate the transcriptional activity of PPAR γ ; however, BADGE can antagonize the ability of agonist ligands such as rosiglitazone to activate the transcriptional and adipogenic action of this receptor. BADGE also specifically blocks the ability of natural adipogenic cell lines such as 3T3-L1 and 3T3-F442A cells to undergo hormone-mediated cell differentiation. These results provide the first pharmacological evidence that PPAR γ activity is required for the hormonally induced differentiation of adipogenic cells.

Peroxisome proliferator-activated receptor (PPAR)¹ γ is a nuclear hormone receptor that is expressed at highest levels in adipose tissue and lower levels in several other tissues. PPAR γ is a major coordinator of adipocyte gene expression and differentiation (1). The expression of this receptor occurs early during the differentiation of preadipocytes, and it is expressed in a highly adipose-selective manner.

PPAR γ has been considered an orphan member of the nuclear hormone receptor superfamily, because no high affinity endogenous ligand has been identified for this receptor. However, a number of synthetic compounds have been shown to bind and activate PPAR γ including a relatively new class of antidiabetic drugs, the thiazolidinediones (2). Thiazolidinediones (TZD) can ameliorate glucose metabolism and improve whole body insulin sensitivity in many animal models of obesity and diabetes. One TZD, troglitazone (RezulinTM), is cur-

rently used in the treatment of Type II diabetes in humans, and a second, rosiglitazone (AvandiaTM), was recently approved by the United States Food and Drug Administration. In addition to synthetic ligands, a number of natural ligands have been described for PPAR γ that include primarily fatty acids and their metabolites (3–5). These ligands, however, have relatively low affinities with $K_d \approx 2$ –50 μ M, and hence it is possible that, analogous to other nuclear hormone receptors, a higher affinity ligand for PPAR γ might exist.

The evidence supporting a key role for PPAR γ in adipogenesis is strong, but it is entirely based on “gain of function” experiments. For example, it has been shown that the ectopic expression and activation of PPAR γ in undifferentiated fibroblasts are sufficient to induce an adipogenic response that includes morphological changes, lipid accumulation, and expression of most of the genes characteristic of this cell type (6). However, until now no experiments have addressed whether PPAR γ function is required for adipocyte differentiation. During a screen for endogenous ligands of PPAR γ we purified and characterized a compound that exhibited PPAR γ binding activity. High pressure liquid and gas chromatography/mass spectrometry (LC/MS/MS and GC/MS, respectively)-based analyses identified this active component as bisphenol A diglycidyl ether (BADGE), a synthetic substance used in the production of polycarbonate and industrial plastics. Competition radioligand binding studies showed this compound to be a ligand for PPAR γ with micromolar affinity. Functional studies indicate that BADGE is a pure antagonist for this receptor.

EXPERIMENTAL PROCEDURES

Cell Culture—3T3-L1 and 3T3-F442A preadipocytes were cultured in Dulbecco's modified Eagle's medium plus 10% serum under non-differentiating and differentiating conditions as described previously (7). Bisphenol A diglycidyl ether (Fluka, Milwaukee, WI) or vehicle (EtOH) was added 24 h prior to induction of differentiation. Medium was replenished with ligands every 2 days. Adipogenesis was determined by staining of lipids with Oil Red O and by the expression of adipocyte-specific RNA markers (8). RNA was isolated using TrizolTM reagent (Life Technologies, Inc.).

Solid Phase Extraction of Cell Nuclei—3T3-L1 and 3T3-F442A cells were lysed in a hypotonic solution containing 1 mM NaHCO₃, 2 mM CaCl₂, and 5 mM MgCl₂. Nuclei were isolated by centrifugation through a 30% sucrose cushion. The nuclear fractions were pooled, resuspended in water, and acidified to a pH value of 3.5 for solid phase extraction. Sep-Pak C₁₈ solid phase extraction cartridges (Waters, Milford, MA) were activated by passing methanol (20 ml) and then water (20 ml) through the stationary phase. Following sample loading, the cartridges were washed with water (20 ml) and then hexane (10 ml), methyl formate (10 ml), and methanol (10 ml) were used to elute the cartridge (9). The extracted fractions were taken to dryness under a gentle stream of nitrogen for radioligand binding analysis. The methyl formate fraction, which carried PPAR γ binding activity, was further purified by HPLC for GC/MS and LC/MS/MS analyses.

* This work was supported by a grant from the Sumitomo Chemical Company (to B. M. S.) and by National Institutes of Health Grant GM38765 (to C. N. S.). The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

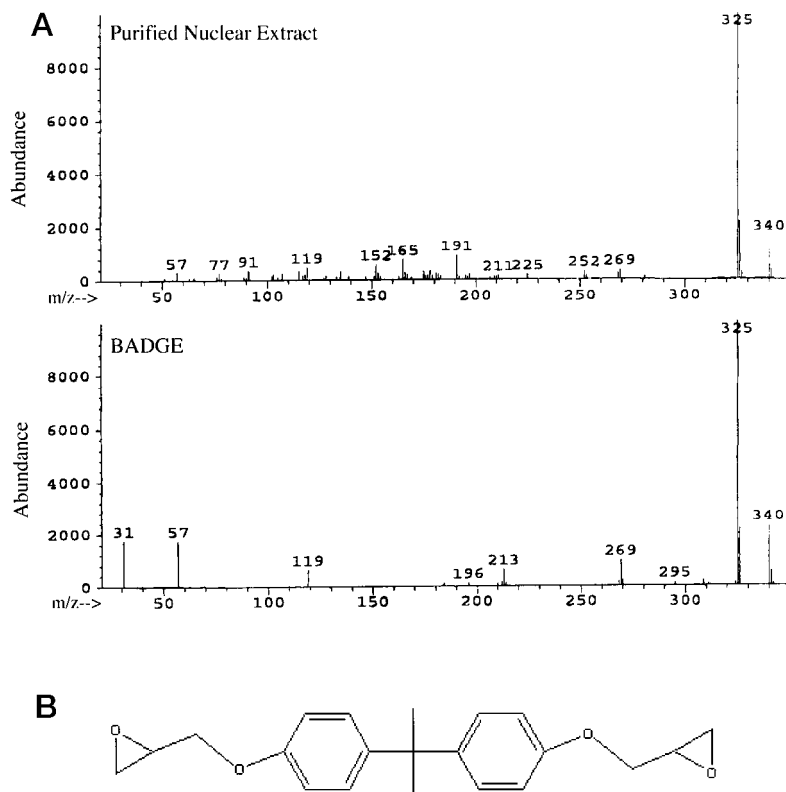
§ Supported by a post-doctoral National Research Service Award from the National Institutes of Health.

** Supported by a post-doctoral fellowship from the Deutsche Forschungsgemeinschaft.

‡‡ To whom correspondence should be addressed: Dana-Farber Cancer Inst., 1 Jimmy Fund Way, Smith 958, Boston, MA 02115. Tel.: 617-632-3587; Fax: 617-632-4655; E-mail: bruce_spiegelman@dfci.harvard.edu.

¹ The abbreviations used are: PPAR, proliferator-activated receptor; TZD, thiazolidinedione; LC, liquid chromatography; GC, gas chromatography; MS, mass spectrometry; BADGE, bisphenol A diglycidyl ether; HPLC, high pressure liquid chromatography; RXR, retinoid X receptor; GR, glucocorticoid receptor; Dex, dexamethasone; IBMX, 3-isobutyl-1-methylxanthine; Ins, insulin.

FIG. 1. GC/MS spectrum and structure of BADGE. A, GC/MS spectrum confirming the identification of BADGE. The upper panel denotes the mass spectrum of an injected sample from HPLC-purified cell nuclear extracts. The lower panel is the mass spectrum of BADGE (base peak of M_r 340) as found in the Wiley Registry of Mass Spectral Data. B, structure of BADGE.



LC/MS/MS and GC/MS Analyses—LC/MS/MS data were acquired with an LCQ (Finnigan, San Jose, CA) quadrupole ion trap mass spectrometer system equipped with an electrospray atmospheric pressure ionization probe. Samples were suspended in mobile phase for injection into the HPLC component, which consisted of a SpectraSYSTEM P4000 (Thermo Separation Products, San Jose, CA) quaternary gradient pump, a Prodigy octadecylsilane-3 (250 $\times$ 2 mm, 5 μ m) column (Phenomenex, Torrance, CA) or a LUNA C18-2 (150 $\times$ 2 mm, 5 μ m) column, and a rapid spectra-scanning SpectraSYSTEM UV2000 (Thermo Separation Products, San Jose, CA) UV-visible absorbance detector. The column was eluted at 0.2 ml/min either isocratically only with methanol/water/acetic acid (69.99:30:0.01, v/v/v) or isocratically for 20 min followed by a linear gradient to 99.99:0.01 methanol/acetic acid (v/v) over 20 min. MS data were collected in the positive ion mode, with the spray voltage set to 5 kV and the capillary to -11 V and 250 $^{\circ}$ C. The scan cycle consisted of a full scan spectrum (MS) acquired with 3 microscans and a 200-ms maximum ion time followed by a product ion mass spectrum (MS/MS) of the most intense full scan MS ion, using a collision energy setting of 30 and with 5 microscans. GC/MS data were acquired with an HP 5890 gas chromatograph (Hewlett Packard, Wilmington, DE) equipped with a DB-1 column (0.25 mm $\times$ 30 m, 0.25 μ m) (J&W Scientific, Folsom, CA) and an HP 5972 mass spectrometer. GC parameters were as follows: injector, 5 μ L, splitless, 300 $^{\circ}$ C; column temperature, initial 100 $^{\circ}$ C, ramp 6 $^{\circ}$ C/min, final 280 $^{\circ}$ C at 40 min. GC/MS spectra were searched utilizing the Wiley Registry of Mass Spectral Data (Palisade Corporation, Newfield, NY).

Ligand Binding Assay for PPAR γ —Ligand binding assays were performed as described previously (4) with some modification. Briefly, His-human PPAR γ ligand-binding domain protein (amino acids 176–477) was expressed in BL21(DE3)pLysS bacteria and partially purified under non-denaturing conditions using Ni-NTA-agarose beads (Qiagen, Valencia, CA). Competition binding assays were performed with [3 H]rosiglitazone (specific activity 50 Ci/mmol, American Radiolabeled Chemicals, St. Louis, MO) and 5 μ L of beads, with and without unlabeled competitor in a ligand binding buffer containing 10 mM Tris (pH 7.4), 50 mM KCl, and 10 mM β -mercaptoethanol. After incubation at 4 $^{\circ}$ C for 2 h, beads were washed with buffer three times to remove unbound ligand. Radioactivity was quantitated by liquid scintillation spectroscopy.

Transfections—NIH-3T3 cells (ATCC, Manassas, VA) grown in 24-well cell culture plates were transfected with pSV-Sport plasmids (500 ng each) encoding PPAR γ , RXR α , DR-1 luciferase, and β -galactosidase utilizing SuperfectTM transfection reagent (Qiagen, Valencia, CA). A fusion protein containing the yeast GAL4 DNA-binding domain linked to the ligand-binding domains of PPAR γ , PPAR α , PPAR δ , and RXR α

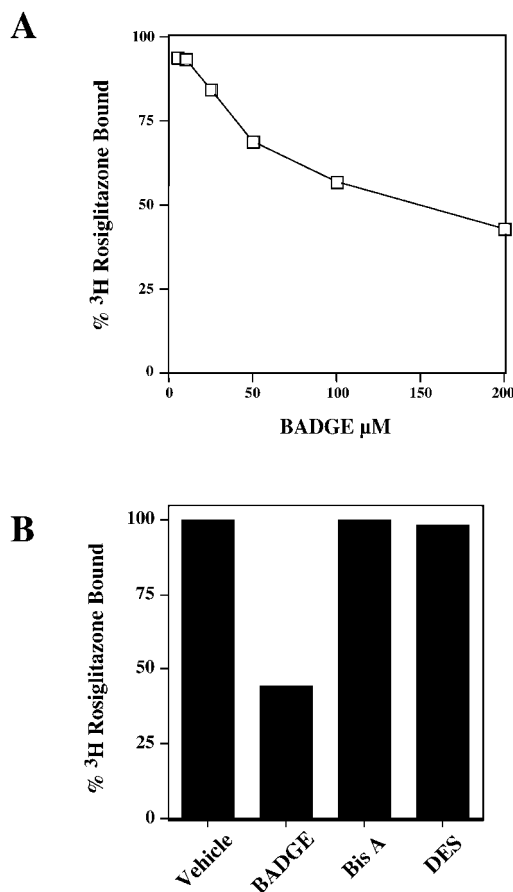
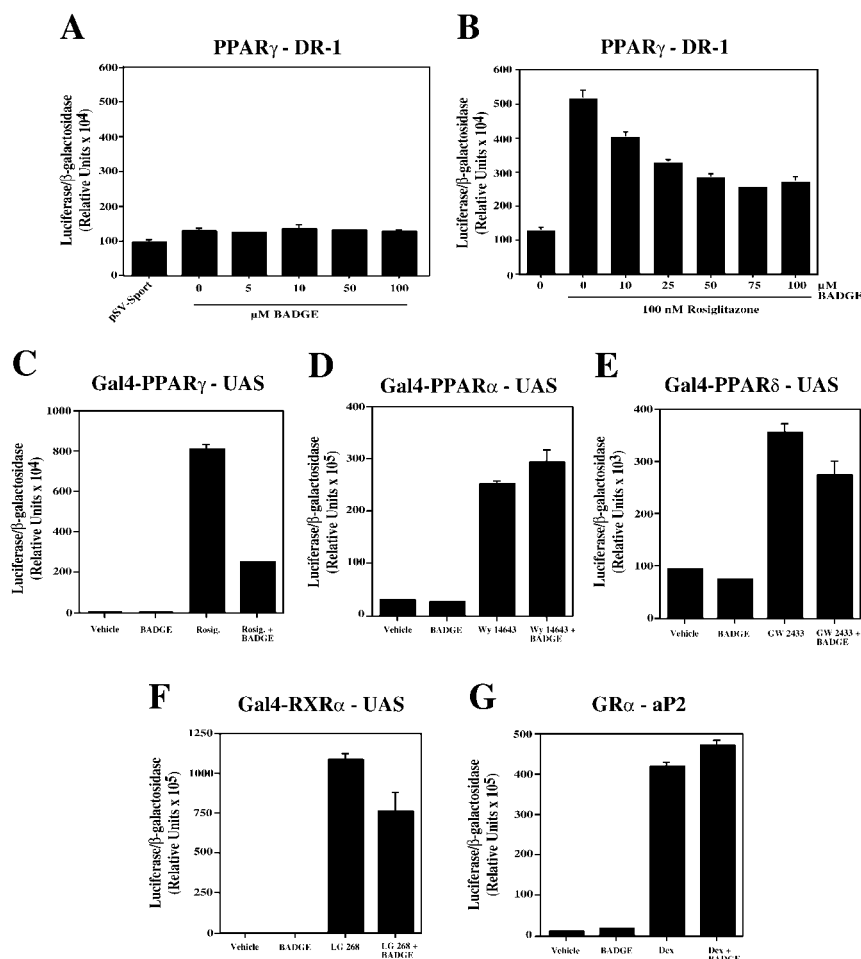


FIG. 2. Binding of BADGE to PPAR γ . A, representative competition binding assay using 5 nM [3 H]rosiglitazone and increasing concentrations of BADGE. The assay was repeated at least three times with similar results. B, competition radioligand binding assay using 10 nM [3 H]rosiglitazone and 100 μ M competitor compounds. Bis A, bisphenol A; DES, diethylstilbestrol.

FIG. 3. Effect of BADGE on PPAR γ transcriptional activity. A, effect of BADGE on PPAR γ /RXR α transactivation. B, effect of BADGE on rosiglitazone-induced transactivation of PPAR γ /RXR α . C, effect of BADGE (100 μ M) on rosiglitazone (500 nM)-induced activation of the isolated ligand-binding domain of PPAR γ . Rosig., rosiglitazone. D, effect of BADGE (100 μ M) on Wy 14643 (1 μ M)-induced activation of the ligand-binding domain of PPAR α . E, effect of BADGE (100 μ M) on GW 2433 (1 μ M)-mediated activation of the ligand-binding domain of PPAR δ . F, effect of BADGE (100 μ M) on LG 268 (25 nM)-induced activation of the ligand-binding domain of RXR α . G, effect of BADGE (100 μ M) on Dex (100 nM)-mediated activation of GR α on an aP2 luciferase reporter. Cells were transfected with nuclear hormone receptor constructs and a luciferase reporter and then treated with ligands. Activation is denoted as relative luciferase units/ β -galactosidase activity.



was also used in transfection experiments with a reporter construct (thymidine kinase-luciferase) containing four copies of a GAL4 upstream activating sequence. Cells were also transfected with a pCMX plasmid encoding full-length human glucocorticoid receptor (GR) α and the aP2 promoter linked to luciferase. Cells were exposed to ligands for 24 h, lysed, and assayed for luciferase and β -galactosidase activity using a 96-well luminometer and a spectrophotometer. Transfections were performed in triplicate.

RESULTS

BADGE Is a PPAR γ Antagonist—During the characterization of PPAR γ ligands derived from cultured adipocytes, we identified BADGE (CAS 1675-54-3), a synthetic compound with industrial applications, namely as a component of epoxy resins (10). BADGE was isolated from nuclear extracts of both 3T3-L1 and 3T3-F442A preadipocytes that were given a differentiation-inducing stimulus. Purification of the compound was first achieved via solid phase extraction of nuclear extracts, followed by reverse phase HPLC. We then performed LC/MS/MS and GC/MS separation of fractions that exhibited activity in a radioligand binding assay, utilizing the bacterially expressed ligand-binding domain of PPAR γ . The identification of BADGE was accomplished using both LC/MS/MS (data not shown) and GC/MS-based analyses. Fig. 1A shows the GC/MS profile comparing an active PPAR γ binding fraction purified by HPLC from 3T3-L1 cells (*upper panel*) with that of BADGE (*lower panel*). Clearly, the GC/MS analysis of the activity purified from the nuclear extract showed a similar MS profile to that of BADGE, with each denoting a base peak of 340, which is the molecular weight of BADGE. It remains to be determined how BADGE accumulated in the differentiating cells; a likely possibility is that it leached from the plastic culture dishes and accumulated in the lipids of the differentiating cells.

BADGE is a ligand for PPAR γ , as can be seen in a radioligand displacement assay utilizing a commercially available preparation (Fig. 2A). 50% displacement of rosiglitazone was achieved at approximately 100 μ M BADGE. Further displacement could not be achieved, probably because of the fact that this represents the solubility limit of this compound in aqueous solution. Nevertheless, the binding of BADGE was selective in that structurally related compounds such as the xenoestrogens bisphenol A and diethylstilbestrol could not displace rosiglitazone (Fig. 2B).

We next examined the effect of BADGE on the transcriptional activity of PPAR γ utilizing a transcription reporter assay. NIH-3T3 cells were transfected with plasmids encoding full-length PPAR γ , RXR α , β -galactosidase, and a DR-1 luciferase reporter. BADGE treatment failed to activate the PPAR γ /RXR α heterodimer in concentrations as high as 100 μ M, the highest concentration that was fully soluble (Fig. 3A). Because BADGE is a ligand for PPAR γ , we examined the possibility that this compound could serve as a receptor antagonist. NIH-3T3 cells were transfected with PPAR γ /RXR α as above and then treated with 100 nM rosiglitazone in the presence or absence of increasing concentrations of BADGE. In this instance, BADGE showed a dose-dependent attenuation of rosiglitazone-induced transactivation (Fig. 3B), suggesting that BADGE is an antagonist for this receptor.

To determine whether such BADGE inhibition could be working on a site outside the ligand-binding domain, NIH-3T3 cells were transfected with a fusion protein between the GAL4 DNA-binding domain and the ligand-binding domain of PPAR γ . As can be seen in Fig. 3C, an antagonistic effect of BADGE on rosiglitazone-stimulated activation was also ob-

served with this construct, suggesting that the ligand-binding domain is the site of BADGE action. Lastly, we wished to determine whether the inhibition of PPAR γ by BADGE was selective for this receptor. NIH-3T3 cells were transfected with plasmids encoding the GAL4 DNA-binding domain and the ligand-binding domain of PPAR α , PPAR δ , or RXR α or a plasmid encoding the full-length human glucocorticoid receptor and treated with their respective ligands in the presence or absence of BADGE. As shown in Fig. 3, C-E, BADGE showed selectivity among PPAR family members. The inhibition of PPAR γ by BADGE in this experiment was $\approx 70\%$, whereas PPAR δ was inhibited by $\approx 23\%$ and PPAR α was not inhibited. Furthermore, BADGE was ineffective in attenuating GR-mediated transcriptional activation (Fig. 3G); however, an inhibitory effect of BADGE ($\approx 30\%$) on ligand-induced activation of RXR α was observed (Fig. 3F).

BADGE Inhibits Multiple Models of Adipocyte Differentiation—Utilizing BADGE as an antagonist, we next addressed the key biological question of whether PPAR γ function is required for adipocyte differentiation. First, we examined the effect of BADGE on PPAR γ ligand-induced differentiation, because this type of differentiation is clearly driven by PPAR γ . 3T3-F442A cells were induced to differentiate for 4 days with 25 nM rosiglitazone in the absence or presence of increasing concentrations of BADGE. The cells were then stained with Oil Red O, which is a marker for neutral lipids. As shown in Fig. 4A, treatment with BADGE resulted in reduced lipid accumulation as observed by Oil Red O.

Because BADGE acts as an antagonist for PPAR γ , we next investigated whether BADGE could affect cell differentiation where endogenous mediators are promoting adipogenesis; this experiment would address whether PPAR γ function is required for hormone-mediated adipose cell differentiation. We utilized 3T3-L1 cells, a well characterized model of adipogenesis. For this experiment, cells were pretreated with different concentrations of BADGE or vehicle and then induced to differentiate with medium containing dexamethasone, 3-isobutyl-1-methylxanthine, and insulin (Dex/IBMX/Ins). As was seen with TZD-induced differentiation, BADGE was able to significantly reduce the amount of lipids in Dex/IBMX/Ins-induced 3T3-L1 cells (Fig. 4B). The effect of BADGE was specific in that bisphenol A was ineffective in attenuating differentiation (data not shown). Of course, there is a concern that this inhibition could be due to a toxic effect of BADGE. This did not appear to be the case, because simultaneous treatment of the cells with a saturating dose of rosiglitazone completely reversed the inhibitory effect of a maximal concentration of BADGE (Fig. 4B). The anti-adipogenic effect of BADGE was also evident on the expression of different adipocyte-specific RNA markers. There was a decrease in the expression of glycerol-3-phosphate dehydrogenase, glucose transporter type 4, and adiponin with no significant alteration in the expression of adipocyte fatty acid-binding protein (aP2) (Fig. 4C). It is likely that the inability of BADGE to inhibit aP2 expression is caused by the difficulty in obtaining complete antagonism for PPAR γ . Rosiglitazone administration with BADGE reversed most of the inhibitory effects. The effect of rosiglitazone on the BADGE inhibited expression of adiponin was not readily assessable because this mRNA is susceptible to down-regulation by TZDs. These results together indicate that PPAR γ activity is required for most morphological and molecular events of adipocyte differentiation.

DISCUSSION

This study characterizes a synthetic antagonist for PPAR γ and demonstrates that loss of PPAR γ function can inhibit adipocyte differentiation. This conclusion is based on our findings that BADGE is a ligand for PPAR γ and that this compound can

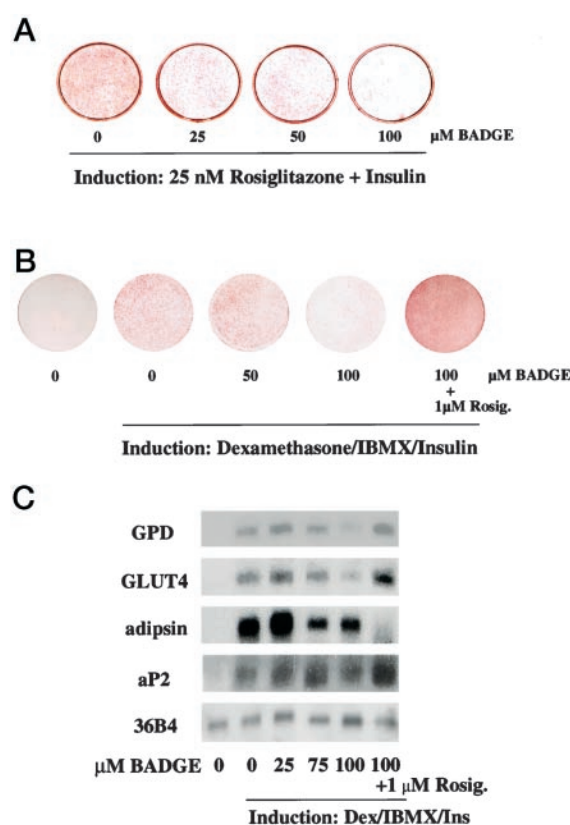


FIG. 4. Effect of BADGE on adipogenesis. A, effect of BADGE on rosiglitazone-induced differentiation of 3T3-F442A cells. Cells were pretreated with BADGE or vehicle (ethanol) for 24 h and then treated with rosiglitazone and insulin (5 μ g/ml) with BADGE or vehicle and with a fresh change of medium every 2 days. After 4 days cells were stained with Oil Red O. B, effect of BADGE on dexamethasone (1 μ M)/IBMX (0.5 mM)/insulin-induced differentiation of 3T3-L1 cells. Cells were pretreated with BADGE or vehicle for 24 h and then induced with Dex/IBMX/Ins with and without BADGE for 2 days. Thereafter, medium was changed to insulin alone with and without BADGE. Cells were stained with Oil Red O after 4 days. Rosig., rosiglitazone. C, effect of BADGE on RNA expression of different adipocyte markers of differentiation. RNA was isolated from 3T3-L1 cells treated under conditions as denoted above. Blots were hybridized with 36B4 cDNA to control for RNA loading. GPD, glycerol-3-phosphate dehydrogenase; GLUT 4, glucose transporter type 4; 36B4, human acid ribosomal phosphoprotein PO.

antagonize the ability of a TZD agonist ligand, rosiglitazone, to stimulate the transcriptional activity of PPAR γ . Furthermore, whereas it was difficult to achieve complete blockage of this receptor because of the relative low affinity and solubility of BADGE, doses could be achieved that interfered with both TZD and hormonally induced adipocyte differentiation. These results are the first pharmacological data to show that PPAR γ function is required for hormonally induced adipocyte differentiation, because evidence supporting a key role for PPAR γ in adipogenesis has been based on gain of function experiments with this receptor. While these studies were being prepared for publication, another group (11) described a high affinity synthetic ligand for PPAR γ (GW0072) that could block TZD-induced adipogenesis in C3H10T1/2 cells. However, this compound was found to be a partial agonist for PPAR γ and thus did not facilitate experiments to determine whether PPAR γ is required in hormone-mediated differentiation.

In addition to regulating adipocyte differentiation, PPAR γ is important in insulin signaling; agonists for PPAR γ are useful in the treatment of patients with Type II diabetes. An antagonist for this receptor could be helpful in investigating the signal transduction pathways involved in PPAR γ -mediated insulin

sensitization, especially in different models of obesity and diabetes. It is also important to note that PPAR γ is expressed in tissues other than fat, such as muscle, colon, and brain. The role of PPAR γ in the development and differentiation of these tissues has not been clearly defined. Thus, it would be of interest to characterize what effect a PPAR γ antagonist would have in non-adipose tissue and even in the whole animal. For instance, it would be interesting to investigate whether a PPAR γ antagonist would be effective in alleviating diet-induced obesity and if so whether such blockage would lead to other deleterious effects such as a reduction in insulin sensitivity. Whereas BADGE itself may not be an appropriate molecule for these studies because of its low affinity, it is possible that higher affinity antagonists can be derived based on this structure.

Another important finding of this study is the identification of BADGE itself as an antagonist of PPAR γ . BADGE and its chemical precursor, bisphenol A, are used commercially in the manufacturing of polycarbonate plastics and have been found in a number of consumer products. For instance, BADGE has been found to migrate into foods from the plastic lining of cans (12) and in susceptors used in microwave cooking (13). BADGE is also present in sealants used in dentistry (14). Because bisphenol A is a known xenoestrogen (15) and endocrine disrupter, it will therefore be important to monitor the possible environmental implications (16) of BADGE exposure in light of its new function as a PPAR γ antagonist.

Acknowledgments—We thank Dr. Steve Kliewer for the bacterial expression vector encoding the ligand-binding domain of hPPAR γ and GW 2433; Dr. Alan Saltiel for rosiglitazone; Dr. Ronald Evans for

GAL4-PPAR δ , GAL4-RXR α , and GR α plasmids; Dr. Richard Heyman for LG 268; and Drs. Bruce Levy, Evan Rosen, and Zhidan Wu for scientific discussions.

REFERENCES

1. Tontonoz, P., Hu, E., Graves, R. A., Budavari, A. I., and Spiegelman, B. M. (1994) *Genes Dev.* **8**, 1224–1234
2. Lehmann, J. M., Moore, L. B., Smith-Oliver, T. A., Wilkison, W. O., Willson, T. M., and Kliewer, S. A. (1995) *J. Biol. Chem.* **270**, 12953–12956
3. Forman, B. M., Tontonoz, P., Chen, J., Brun, R. P., Spiegelman, B. M., and Evans, R. M. (1995) *Cell* **83**, 803–812
4. Kliewer, S. A., Lenhard, J. M., Willson, T. M., Patel, I., Morris, D. C., and Lehmann, J. M. (1995) *Cell* **83**, 813–819
5. Kliewer, S. A., Sundseth, S. S., Jones, S. A., Brown, P. J., Wisely, G. B., Koble, C. S., Devchand, P., Wahli, W., Willson, T. M., Lenhard, J. M., and Lehmann, J. M. (1997) *Proc. Natl. Acad. Sci. U. S. A.* **94**, 4318–4323
6. Tontonoz, P., Hu, E., and Spiegelman, B. M. (1994) *Cell* **79**, 1147–1156
7. Cook, K. S., Hunt, C. R., and Spiegelman, B. M. (1985) *J. Cell Biol.* **100**, 514–520
8. Tontonoz, P., Hu, E., and Spiegelman, B. M. (1995) *Curr. Opin. Genet. Dev.* **5**, 571–576
9. Serhan, C. N., and Sheppard, K. A. (1990) *J. Clin. Invest.* **85**, 772–780
10. Ben-Jonathan, N., and Steinmetz, R. (1998) *Trends Endocrinol. Metab.* **9**, 124–128
11. Oberfield, J. L., Collins, J. L., Holmes, C. P., Goreham, D. M., Cooper, J. P., Cobb, J. E., Lenhard, J. M., Hull-Ryde, E. A., Mohr, C. P., Blanchard, S. G., Parks, D. J., Moore, L. B., Lehmann, J. M., Plunket, K., Miller, A. B., Milburn, M. V., Kliewer, S. A., and Willson, T. M. (1999) *Proc. Natl. Acad. Sci. U. S. A.* **96**, 6102–6106
12. Summerfield, W., Goodson, A., and Cooper, I. (1998) *Food Addit. Contam.* **15**, 818–830
13. Sharman, M., Honeybone, C. A., Jickells, S. M., and Castle, L. (1995) *Food Addit. Contam.* **12**, 779–787
14. Olea, N., Pulgar, R., Perez, P., Olea-Serrano, F., Rivas, A., Novillo-Fertrell, A., Pedraza, V., Soto, A. M., and Sonnenschein, C. (1996) *Environ. Health Perspect.* **104**, 298–305
15. Steinmetz, R., Mitchner, N., Grant, A., Allen, D., Bigsby, R., and Ben-Jonathan, N. (1998) *Endocrinology* **139**, 2741–2747
16. Limbird, L. E., and Taylor, P. (1998) *Cell* **93**, 157–163

A Synthetic Antagonist for the Peroxisome Proliferator-activated Receptor γ Inhibits Adipocyte Differentiation

Harold M. Wright, Clary B. Clish, Toshiyuki Mikami, Stefanie Hauser, Kazunori Yanagi, Ryuji Hiramatsu, Charles N. Serhan and Bruce M. Spiegelman

J. Biol. Chem. 2000, 275:1873-1877.
doi: 10.1074/jbc.275.3.1873

Access the most updated version of this article at <http://www.jbc.org/content/275/3/1873>

Alerts:

- [When this article is cited](#)
- [When a correction for this article is posted](#)

[Click here](#) to choose from all of JBC's e-mail alerts

This article cites 16 references, 5 of which can be accessed free at <http://www.jbc.org/content/275/3/1873.full.html#ref-list-1>